

Post Translational Events

→ Protein Folding →

• Chaperons

Hsp 70, Hsp 90, Hsp 60

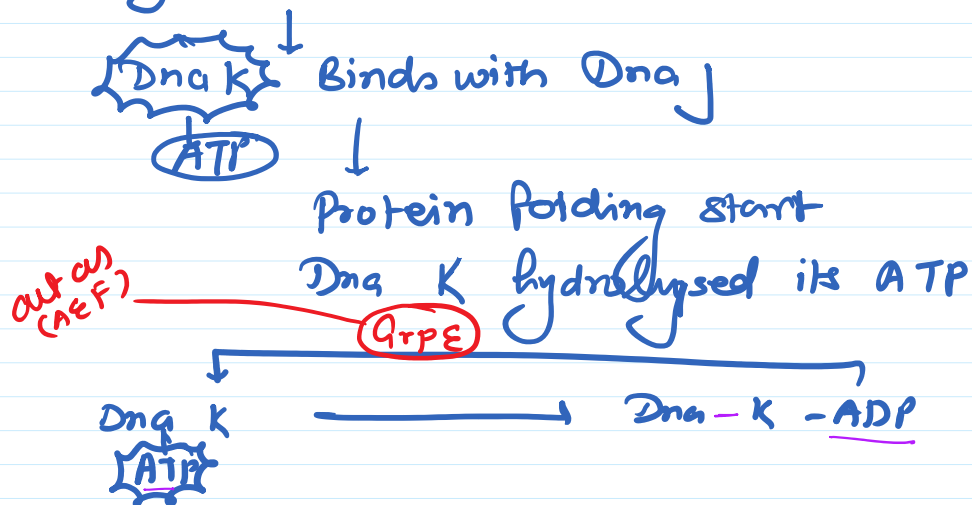
Eubacteria

• Dna K (Hsp 70 family)



• Dna J (Hsp 40 family)

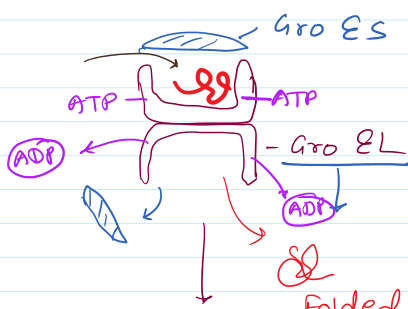
* Dna J associated with unfolded protein



* in Eubacteria - Drum like protein [Chaperon]

are +nt for protein folding
belongs to hsp 60 family

Named: Gro EL & Gro ES




• drum like protein

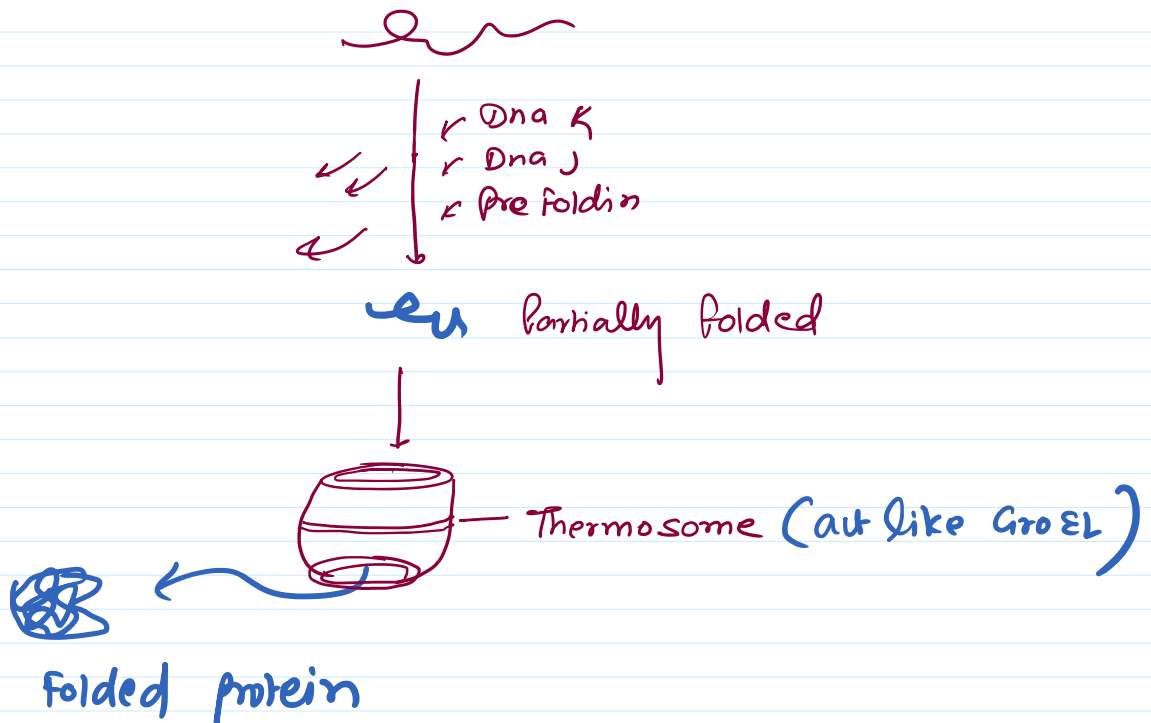
• +nt in Dimer Form

• each Dimer contain 7 subunit

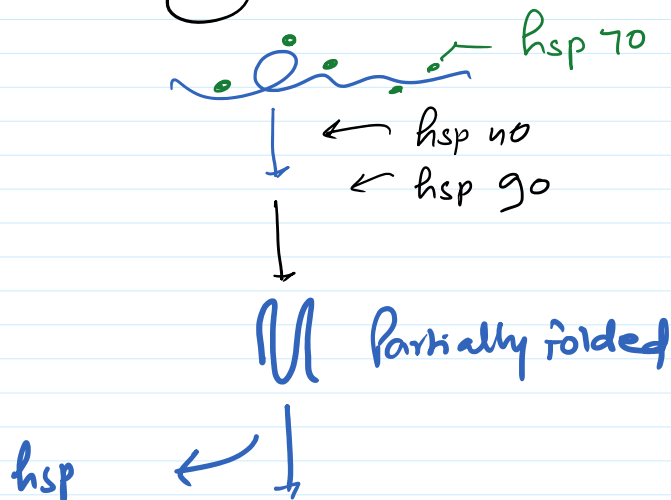
→ Folded protein

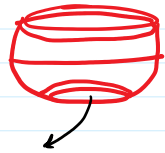
Archae Bacteria -

- Dna K - (hsp 70)
 - Dna J - (hsp 40)
 - Pre-foldin (hsp)
- +  ATP
↓
act as co factor



In Eukaryotic System



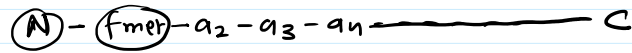


TRIC [8 subunit complex]

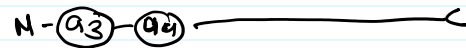
↳ act like Gro EL

folded protein

Post Translational modification -
in prokaryotes -



↓
← deformylase
← aminopeptidase



in Eukaryotes

◦ Proteolytic Cleavage

eg. Cleavage of signal peptide from
certain protein [secretory protein]

↓
Co-Translational Event

* Post Translational Proteolytic Cleavage

◦ In mitochondria - Removal of mTS (matrix Targeting signal)

◦ In chloroplast - " " STS (stromal Targeting signal)

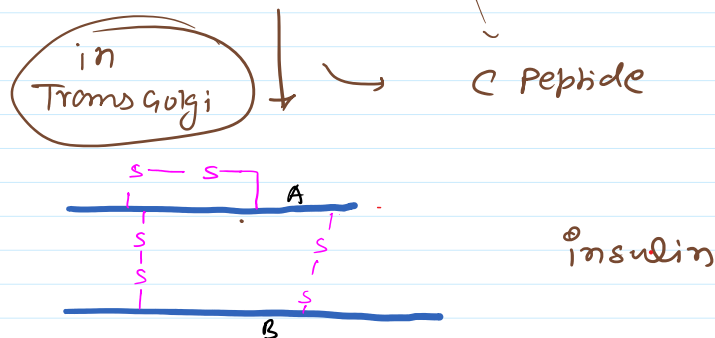
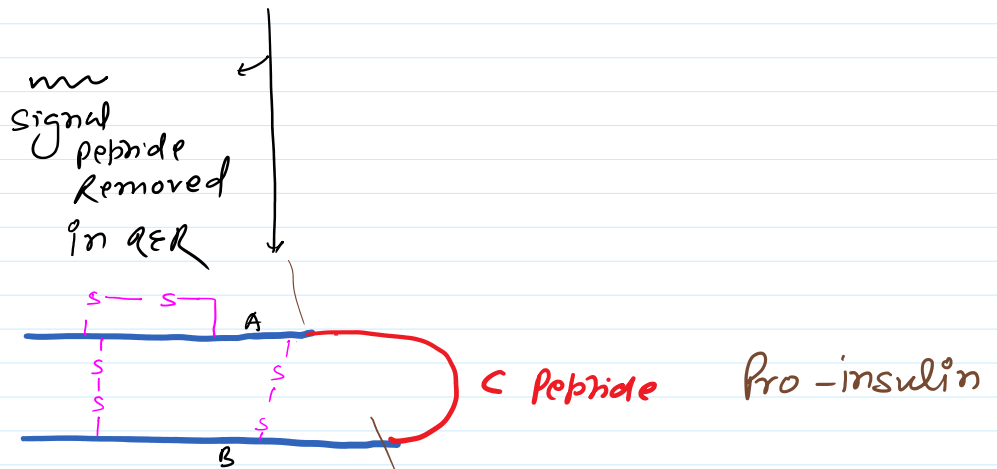
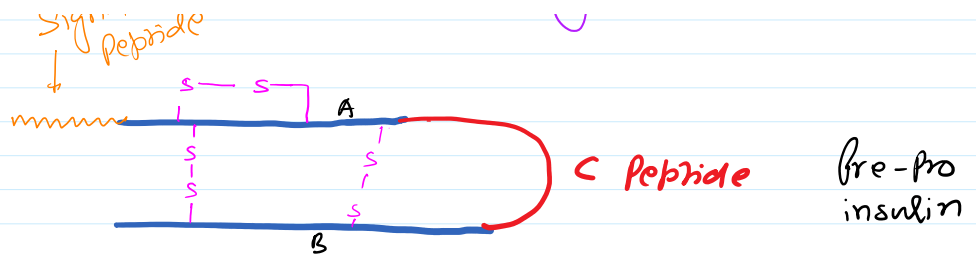
◦ 2 Round proteolytic cleavage in insulin

◦ ① Removal of signal peptide

② Removal of C-peptide

Signal Peptide
↓





modification of protein

Proteolytic Processing

① myristylation [14 C F.A]

- 14 C Fatty acid attached with protein

↓

Co-Translational modification of lipid Anchored protein
eg Gα protein

② Farnesylation [Post Translational modification]

10C - Geranyl

15C - Farnesyl

20C - Geranyl-Geranyl

15C - farnesyl
 20C - Geranyl-Geranyl
 add on C Ter. of Peptide
 eg - Ras protein

③ - Palmitoylation (16 C F.A.)

④ GPI Anchored

⑤ Acetylation on K or R
 Residue of N Ter. of Histone

Non acetylated Histone
 ↓
 Heterochromatin

Acetylated
 ↓
 (Euchromatin)

⑥ Glycosylation

① N-linked Glycosylation → occurs in RER [Co-Translational]

② O-linked → " " Golgi [Post-Translational]

⑦ Phosphorylation

- on α Chain of Ser/Thr/Tyr/His/Asp
- has regulatory role

⑧ Ubiquitination

mono-ubiquitination

→ Regulatory Role

→ involve in Repair mechanism

PCNA - Ub ← DNA Damage

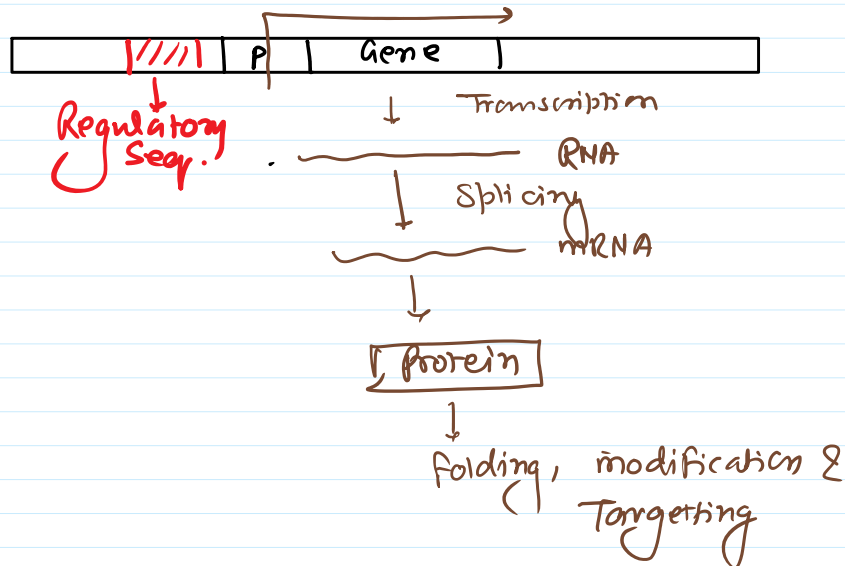
Polyubiquitination

• on K Destruction Box

• Signal for 26-S Proteasomal degradation

↓
Recruit Repair
System

Regulation of Gene Expression



* No. of gene +nt in E. coli = 4400 gene
Yeast = 7000 gene
Human = 27,000 gene

* But all gene are not expressed at a time
Differential gene expression can be seen.

↓
• There must be some regulatory mechanism

→ Regulation of a Gene Expression at
Different level →

① Transcriptional Gene Regulation-

• Initiation level - most common

↳ most preferred because - low energy investment

• Disadvantage - Slow Response

• Post Transcriptional gene Regulation.

ii) Translational Regulation

① • initiation level

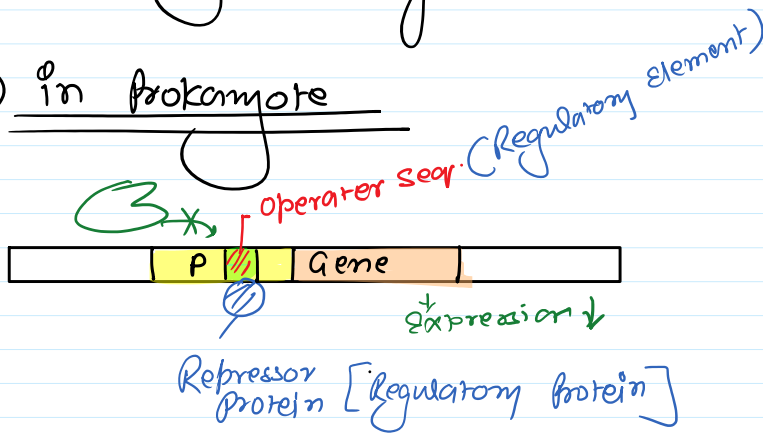
② Post translational Regulation

↓
Advantage - Response fast

Disadvantage - Energy investment ↑

Regulation of Gene Expression

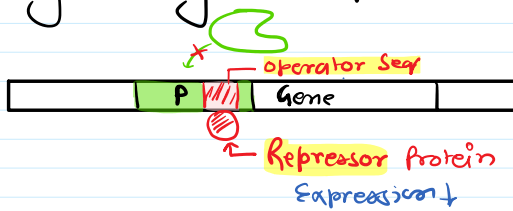
① in prokaryote



• Control of Gene Expression Through Repressor protein is common among prokaryotes

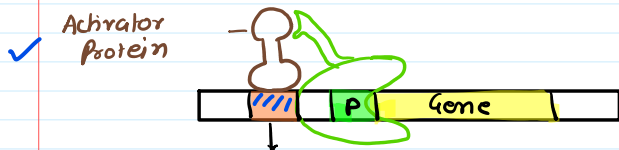
Reason - Prokaryotic Promoter is strong Promoter

Regulation of Gene Expression



- control of gene Expression Through repressor protein is common among prokaryote
- Reason - Prokaryotic gene Promoter is Strong Promoter

Eukaryotic gene Expression -



✓ Enhancer Seq. } → ↑ Expression

- Expression is generally controlled by Activator Protein
- Reason - Promoter is weak

Basal Transcription Rate -

Rate of Transcription - without influence of activator & Repressor

- generally - 1 mRNA/min
- GatA - 10 mRNA/min ✓
- ara b = 100 mRNA/min

Promoter of ara b gene is Strongest among prokaryote

Variation in Transcription Rate is depends upon - Promoter Strength

Strong Promoter = ↑ Basal Transcription Rate
weak " = ↓ " " "

In prokaryote Promoter -

Consensus Seq.

-10	-35	Up Element	Promoter Strength
✓ TATAAT	TTGACA ✓	+nt	Strong ✓
TATAAT	T+GACA	-nt	moderate ✓
TATAAT	T G A G A	-nt	weak

Differential gene Expression [Prokaryote]

Various Sigma factor (σ) are +nt in prokaryote for differential gene expression

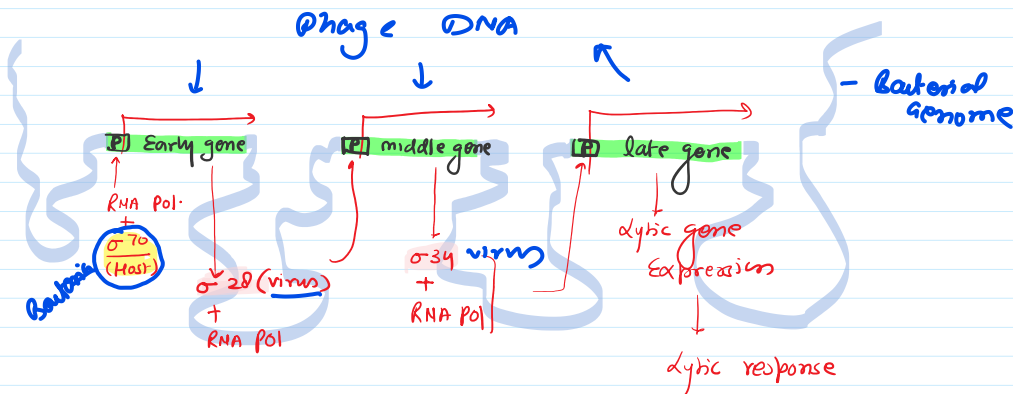
eg - σ^{70} - responsible for Expression of most of gene

~~gmp~~

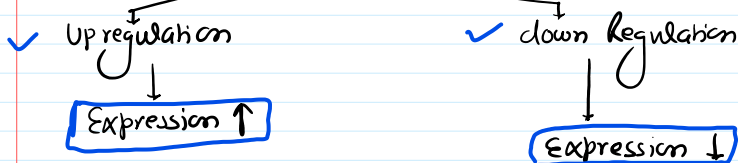
Gene -	
rpo H	σ^{32} - Heat Shock protein Expression
rpo N	σ^{54} - Nif gene
?	σ^S - Sporulation
rpo F	σ^{28} - Flagellar protein Expression
rpo E	σ^{24} - hsp of Periplasm
fec I	σ^{19} - Expression of Iron Transport gene
rpo S	σ^{38} - Stationary Phase gene Expression

Basal gene Expression of Bacteriophage SpoII ✓

Host - Bacillus Subtilis ✓

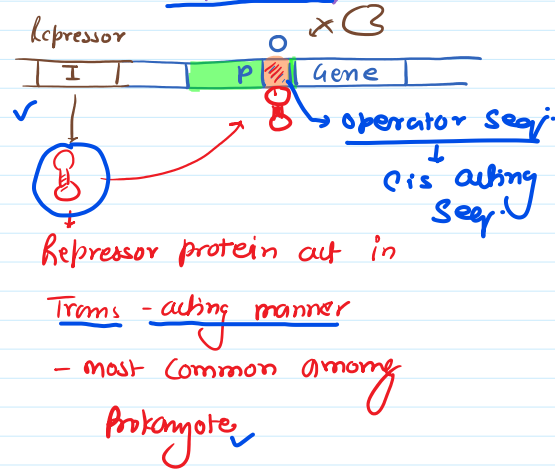
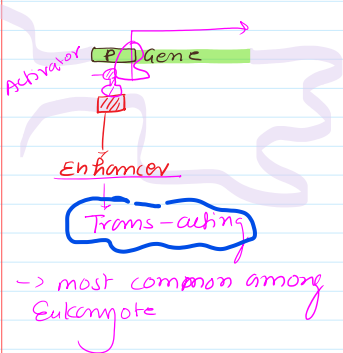
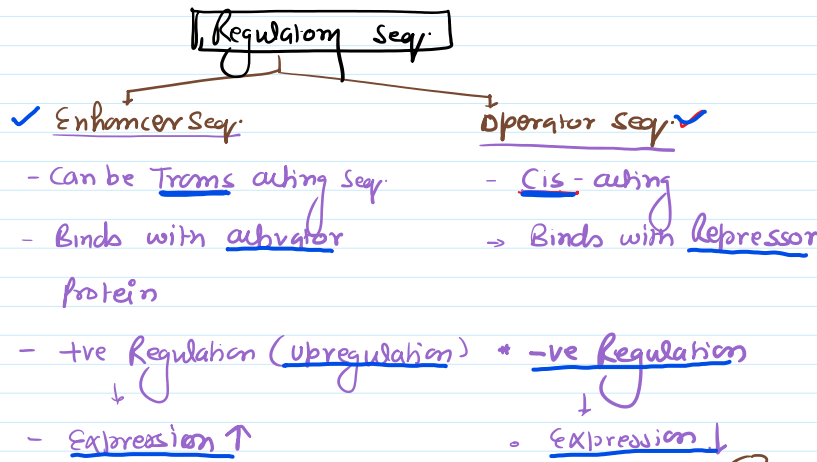


Regulation of Gene Expression

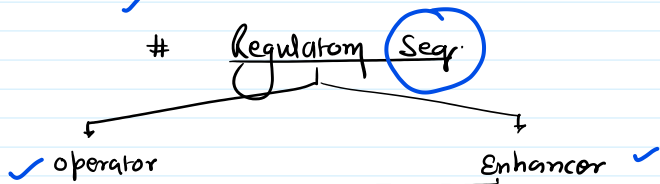


Regulation Seq.

expression ↑



→ Enhancer may be cis acting also



✓ • Bipartite

✓ • inverted

✓ • non-Specific in orientation

But promoter is specific with its orientation

eg - lac Operator



inverted seq.

How Activator Protein Control gene Expression

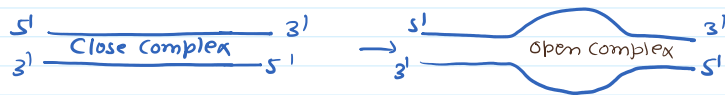
✓ - Induction

✓ - Positive Control of Gene Expression

① Recruitment of RNA Pol.



② Positive Allosteric Change



eg - NtrC

③ Promoter Clearance / Promoter Escape

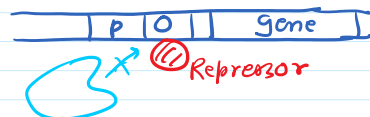
↓
Elongation Start

eg - malT
↓
(maltose)

Repressor protein -

↳ Negative Regulator

① Steric Hindrance



eg - lac operon

② Negative Allosteric Change -

Open Complex → Close Complex

eg - ArpC [Arabinose]

Repressor or Activator protein contain

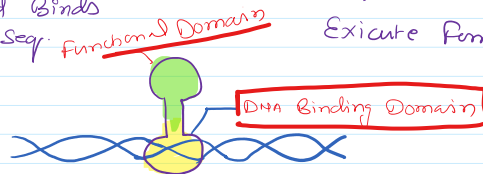
2 Domain -

DNA Binding Domain

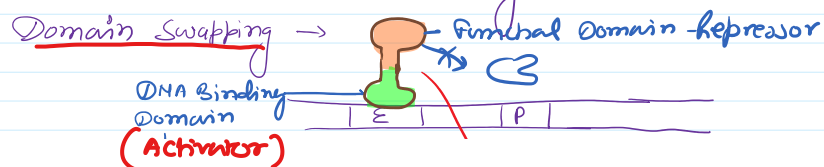
↓
• Recognize and Bind
on DNA Seq.

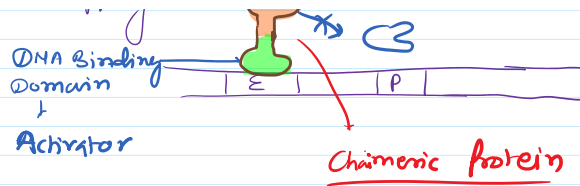
Functional Domain

↓
Execute Function



if activator functional Domain is replaced
with Repressor functional Domain by





DNA Binding Domain

↳ Contain Super 2° Str. that interacts with DNA
K/a motif.

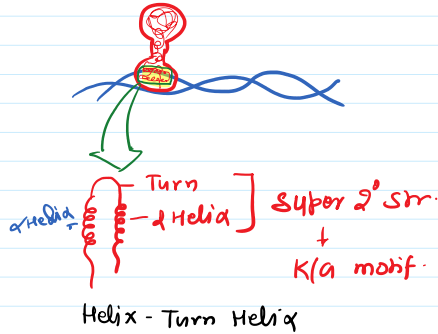
✓ motif

① Helix - Turn - Helix

→ most common in prokaryotes

eg - Lac repressor

- 1 repressor
- Trp repressor
- Lex-A repressor
- CAP activator

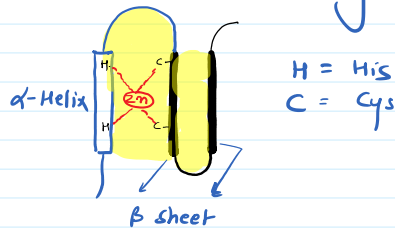


1 α Helix interacts with

DNA & another Helix
stabilize the binding of
that α Helix

② Zinc Finger Motif

- common in Eukaryotes



eg - Transcription factor - III A, III C

Steroid Receptor family [Glucocorticoid Receptor
Retinoic acid Receptor]

③ Basic Zipper

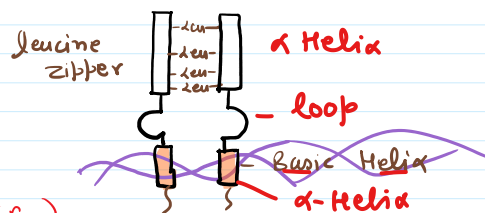
↳ Interacts with DNA

✓ Leucine zipper



eg - fos, Jun (T.F.)

✓ Helix-loop-Helix



eg - myc, myb (T.F.)

gmb

Functional Domain

◦ Activator Domain ✓

- Acidic Domain
- Proline Rich a.a.
- Glutamine Rich a.a.

◦ Repressor Domain

- not generalized

Gene Regulation in Prokaryotes

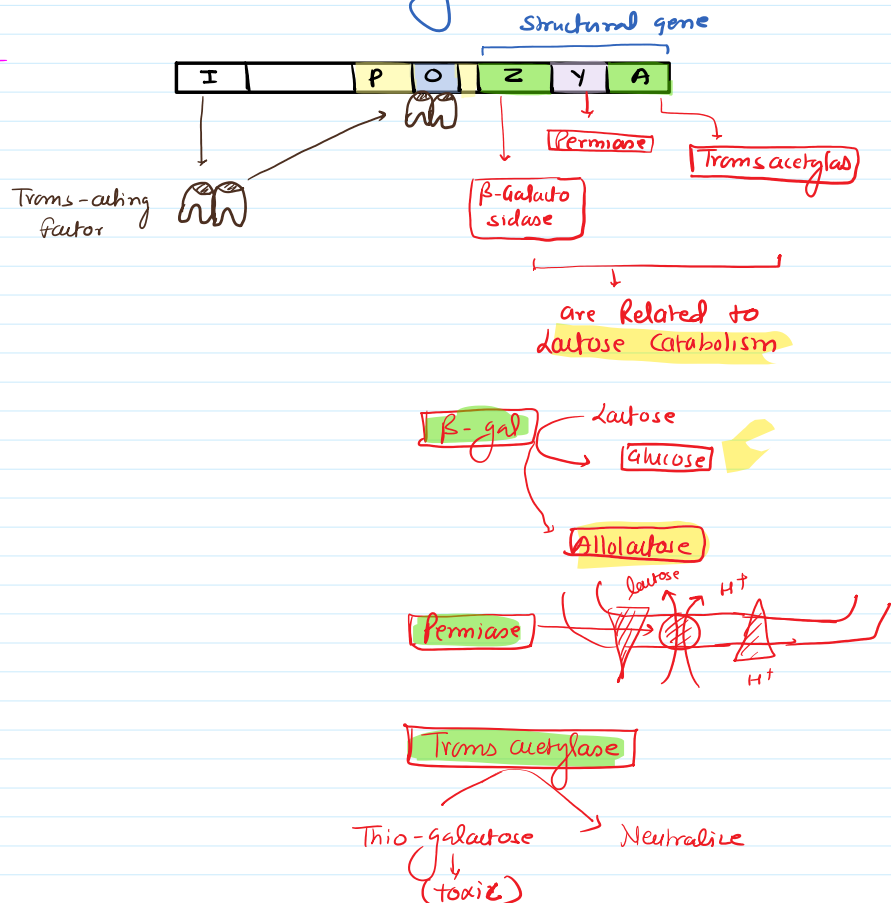
◦ Regulation at Transcription initiation level-

① lac Operon

- Co-ordinated Regulation of gene Expression
in E. coli

- Discovered by Jacob and monod

Condition 1
◦ Lactose - nt
◦ Glucose +nt



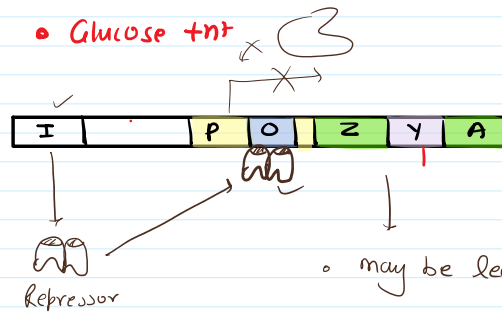
Regulatory Seq. → operator [1-3 Kb]

overlapped in Promoter

Condition 1

★ Lactose - nt

◦ Glucose +nt



◦ may be leaky Transcription occurs

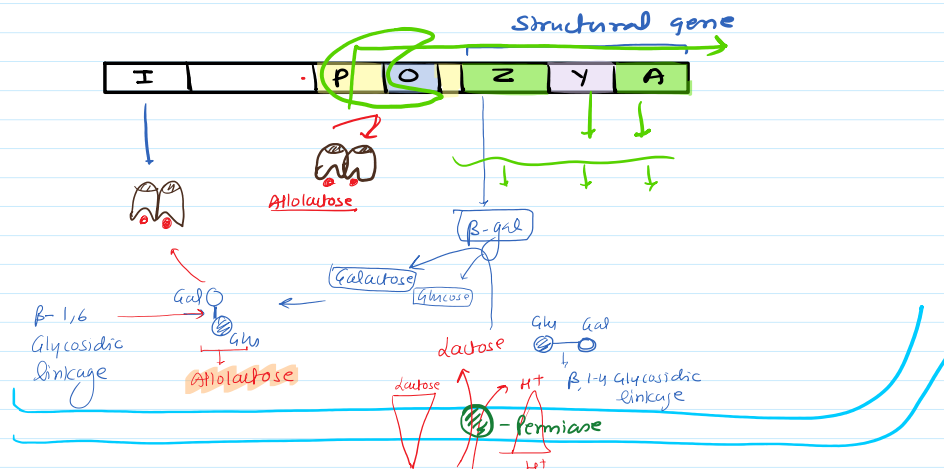
◦ 0-1 Transcript / min

◦ few permease & β Gal may be +nt

Condition 2

◦ Lactose +nt

◦ Glucose -nt



Allolactose has ↑ affinity with Repressor

↓

Allolactose Binds with Repressor & brings the Conformational Change in Repressor

↓

Repressor dissociates from operator

↓

Expression starts

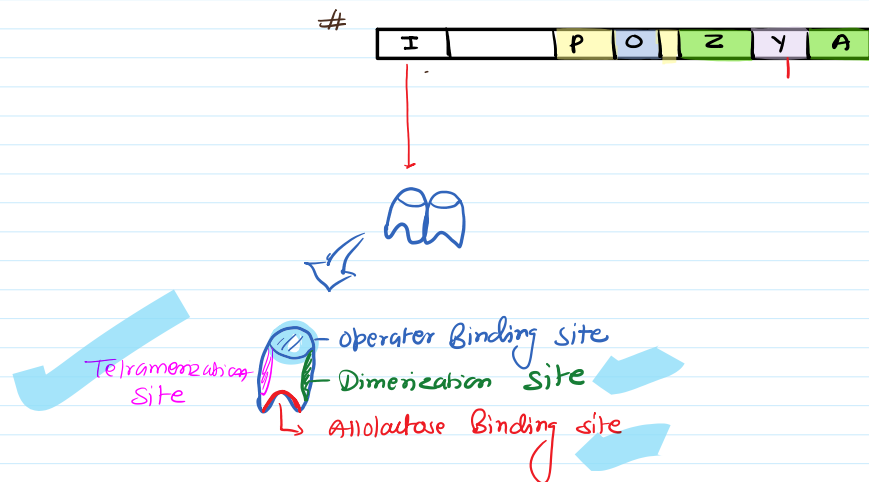
[Allolactose] → Natural Inducer [induce Expression of gene]

→ In Artificial Expression System → IPTG is used as artificial inducer

[IPTG] - Artificial inducer

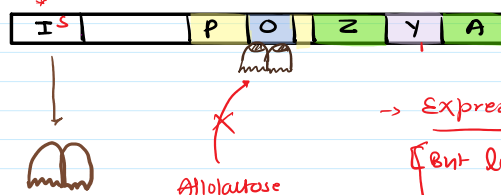
↳ [Isopropyl Thiogalactoside]

- ↳ Small & non-polar
 - ↳ Can cross mem. without need of Transporter
 - ↳ ↑ Affinity with Repressor
 - ↳ analog of allolactose
- IPTG gives at 16°C in Broth Culture for gene Expression



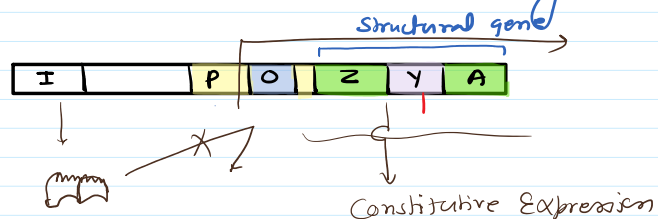
if Allolactose Binding site is mutated
↓
Repressor become Super repressor

Super repressor [Gain of Function]



→ Expression - off
[But leaky Transcription can be seen]

② if loss of function in Repressor
↳ mutation in DNA Binding site



③ if loss of function in operator [O^c]

- ↳ Can not Bind with Repressor
- ↳ Constitutive Expression Can be Seen

Inducer +nt
- - -

Inducer -nt
- - -